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Ant Recruitment to Salt Along Chicago's Urban Gradient

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Abstract

Sodium is vital for maintaining osmotic balance, muscle activity, and nervous system function in many organisms. The availability of sodium can drastically impact populations within ecosystems. Salt content from human foods and salt from winter road treatments could increase Na^+ availability. I hypothesize that Na^+ availability is more limited when moving away from Chicago. I baited ants along two 50km transects extending West and North from downtown Chicago. Transects contained 10 evenly spaced forested sites. Baits were equally split between H_20, 20% sugar, and 1% salt concentration. Thirteen ant species were collected, but only three species recruited to salt baits. There is no relationship between ant recruitment to salt and distance from Chicago. It is possible that proximity to roads is the more important variable in determining salt availability. Thus, the urban gradient may not matter due to the spatial scale in which ants live.

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Ant Recruitment to Salt Along Chicago’s Urban Gradient

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Marco Andres Servin

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The report of the investigation undertaken as a
Senior Thesis, to carry two courses of credit in
the Department of Biology

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Abstract

Sodium is vital for maintaining osmotic balance, muscle activity, and nervous system function in many organisms. The availability of sodium can drastically impact populations within ecosystems. Salt content from human foods and salt from winter road treatments could increase Na^+ availability. I hypothesize that Na^+ availability is more limited when moving away from Chicago. I baited ants along two 50km transects extending West and North from downtown Chicago. Transects contained 10 evenly spaced forested sites. Baits were equally split between H_20, 20% sugar, and 1% salt concentration. Thirteen ant species were collected, but only three species recruited to salt baits. There is no relationship between ant recruitment to salt and distance from Chicago. It is possible that proximity to roads is the more important variable in determining salt availability. Thus, the urban gradient may not matter due to the spatial scale in which ants live.

for

Sean B Menke

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Chapter 1 – Sodium Where to Find it and Why It's Important

Introduction to Salts

All organisms on earth require nutrients in the form of elements to survive; many times, homeostatic mechanisms function to drive an organism to acquire these elements. Sodium, Na, is one of these common elements. Usually, sodium is found as salt in Earth's oceans and terrestrial salt deposits. While organisms require some level of sodium, too much can over saturate cells within the organisms, causing a variety of stresses. Recent studies have shown increasing anthropogenic effects causing greater salt and salinity concentrations in the natural environment (Banin & Fish 1995). Exploring salt's importance, availability, and potential consequences on organisms are vital to maintaining our increasingly urban landscape.

Salts are a classification of minerals made from a reaction between an acid and a base through cations and anions. The most commonly known salt is sodium chloride (NaCl), but other elements or compounds such as, magnesium (Mg), calcium (Ca), carbonate (CO^{2-}), nitrate (NO_{3-}), and sulfate (SO_4^{2-}), can all form salts (Britannica 2019). Earth's natural salt sources are ocean water and natural salt deposits. Of all of Earth's water, 97% is saltwater, leaving 3% as freshwater; approximately 1% of which is readily available for our use (N.A.S.A. 2020). However, on land salt is much rarer.

On the Earth's crust, salt is commonly found as the mineral halite, known as rock salt (Bauholz 2017). Halite can be found around salt springs, salt lakes, and in the ocean. It can also be found in salt domes and provide important traps for oil deposits (Najiya & Cole 2020). On the surface, halite deposits form by the slow evaporation and eventual drying of enclosed bodies of saltwater. Buried salt deposits are commonly found in sedimentary rock beds and in saline lake deposits such as the Great Salt Lake in Utah (Arnow & Stephens 1990). Often, organisms are not interested in the mineral form of salt but in its salinity once it has dissolved into water or soil.

Sodium: From Small to Big Effects

Salts are needed by all organisms in varying concentrations, with some salts containing sodium. Sodium is not only an essential building block during development but is crucial for maintenance within cells as well. Sodium deficiency for organisms or their ecosystems can have dire consequences on their ability to survive. The sodium required by many organisms is a non-metal element that can flow between cells. Sodium (Na) is a cation usually found as Na^+. Na^+ acts as a key ion in extracellular space, and it can enter cells through a variety of routes, including permeation and voltage-gated cation channels (Yu et al. 2010). Organisms must be able to handle the outside environment while maintaining their interior systems. When their systems are at risk of being exceeded, many organisms have mechanisms to return the systems to a safe state of function. The same homeostatic mechanisms regulate chemical balances such as salt, and metabolic conditions (Fraústo da Silva & Williams 2001). Thus, it is important for organisms within ecosystems to have salt accessible, yet not in supersaturated amounts, to allow organisms to maintain appropriate sodium levels.

Organismal cells require sodium because it plays an important role in animal physiology. The concentration of Na^+ ions inside cells is generally lower than the extracellular fluid surrounding them. This sodium ion gradient is partially counterbalanced by an opposite potassium ion (K^+) gradient. Cell membranes contain an enzyme, Na+/K+-ATPase that acts as a sodium pump. This "sodium pump" maintains the gradients of both Na^+ and K^+ ions across a cellular membrane (Pirahanchi & Aeddula 2019). The positive ions also aid in creating an electrical potential difference, or voltage. The voltage within these gradients is crucial for nerve transmission and muscle contraction.

Sodium ion gradients are also responsible for various transport processes, such as sugar transport in the intestine and amino acid transport in red blood cells (Bentley 2015). Organisms, especially herbivores and microbial decomposers, often struggle to maintain their sodium levels (Kaspari et al. 2008). In humans, sodium is lost along with water, when we perspire. This is why it is important to take in salt as well as water when exercising, to avoid dehydration and maintain the correct composition of body fluids (Bentley 2015). Studies have shown that Na+ influx into cells is accompanied by Cl^- ions and water, which can lead to acute neuronal swelling and damage in excess amounts (Yu et al. 2010). Moreover, Na^+ entry via Na^+/H^+ exchange can change intracellular pH, and thus regulate many cellular functions including enzyme activity, neuronal growth, and cell death through apoptosis. (Yu et al. 2010).

While Na+ is a necessity of organisms, it can be detrimental in excess amounts. Therefore, it is important to acknowledge that every substance has a limit before it becomes toxic. When sodium begins to exceed the cells' threshold, the Na^+ ion can begin to cause harm. When appearing suddenly at high concentrations, Na^+ can produce osmotic consequences that disrupt cell membranes and inhibit growth (Kronzucker et al. 2013). Animals can consume plants with a supersaturated amount of sodium, who then are taking in the plant's sodium content. Often, research is not only done on an organism's Na^+ and salts, but on salinity within an environment.

Salinity refers to the presence of salts in solutions or mixtures; in the natural environment scientists are concerned with its affects in water and soil, as both are recourses used and lived in by organisms (Corwin & Yemeto 2017). Thus, measuring the salinity in solutions is vital for predicting how salts will affect other organisms and environments. For example, Na+ is more likely to be absorbed into the soil than Ca^{2+} when they are dissolved from their ionic bonds (Wong et al. 2008). The salinity of water is usually expressed in terms of electrical conductivity (EC) and total dissolved solids (TDS). Electrical conductivity (EC) refers to a substance's ability to conduct electricity through the transfer of electrons and is the parameter often used to express salinity; higher EC indicates greater salinity. Total dissolved solids (TDS) express the mass of dissolved ions and molecules per unit volume of water and can be used to express the salinity of water; higher salt levels in solutions produce higher TDS readings (Corwin & Yemeto 2017; Montazar 2020). The water we use for drinking, irrigation, and recreation depends on the type and concentration of dissolved salts in water, all of which can lead to increasing salinity in the natural environment.

Variation in salinity occurs naturally along coastal areas, including atmospheric deposits of oceanic salts and seawater intrusion into groundwater basins (Montazar 2020; Kaspari et al. 2008). Oceanic aerosols carry salt and deposit it towards the interior of continents; the rate of deposited salt decreases with distance from the coasts (Kaspari et al. 2008). Salinity levels found in soils are caused by rising ground waters, inland saline lakes, and leaching of saline lands (California 2019; Montazar 2020). While the ocean provides a primary source of natural salt deposits, industries commonly extract salt directly from the surface, ground waters, and mines. The water-based sources contain dissolved salts with varying concentrations, depending on the salt, the substrate, and the history of the water itself (California 2019; Montazar 2020). There are sources of salts caused by human activity such as, irrigation, drainage water, chemical fertilizers, animal wastes, sewage sludges, and oil and gas field brines. Researchers are constantly measuring the salinity levels in various of the previously mentioned fields to assure a safe level of salt in our environment.

Salts and Sodium in Urban Environments

The United Nations reports that more than half the world's population lives in urban cities (United Nations Statistics Division 2020.) Studies have shown that urban land use significantly contributes to changes in the environment (Grimm et al. 2008). Increasing urbanization of previously natural environments means that people are encountering more wildlife species in urban areas (Shochat et al. 2006). While many people are aware the world is changing due to climate change, they forget anthropogenic activities play a key role on environmental change. Our society needs undisrupted infrastructure to move goods and people in an efficient manner. For many of the Northern and Midwest states, snowfall and icy roads can drastically affect road systems. One anthropogenic source of salts is the introduction of salt melts along road systems (Kelly et al. 2019).

The cheapest, quickest, and most effective treatment to cope with winter ice and snow is applying salt melts to roads and highways; specifically, in the form of NaCl (Salt Institute 2004). Other chloride salt compounds have been used instead of NaCl. Calcium chloride and magnesium chloride are used to melt snow and ice more quickly at lower temperatures but are seven times more expensive than the sodium chloride. The addition of organic alternatives can impose different environmental stress by introducing other ions into the environment (Dugan et al. 2017).

As urbanization expands, researchers need to monitor the salts being put into the environment. Regions that heavily rely on salts will only be putting more salts into the environment as they prioritize road safety and efficiency. As stated previously, placing salts into the environment also means that the ionic bonds holding the elements together will break, allowing the detached ions to be displaced. Some researchers (Asefa et al. 2005; Rengasamy 2010) have been monitoring the salinity through environments and time, however, not many have explored the long-term effects of placing more Na^+ and Cl^- ions into the environment.

Chapter 2 – Ant Recruitment to Salt Across Chicago

Introduction

As anthropomorphic activity continues to change urban landscapes, it is crucial for scientists to investigate urban biomes and their potential effects on organisms. The future of organisms in urban areas may depend on finding information on species that play vital ecological roles in the environment. A variety of animals in urban areas have the ability to adapt to changing environmental conditions (Magle & Angeloni 2011; Magle et al. 2015). Magle et al. (2015) observed that housing density had varying impacts on coyotes, racoons, and opossums; increasing housing density had a negative relationship with colonization probability for coyotes and racoons, while having a positive correlation for opossums. Magle & Angeloni (2011) have also found that isolated urban colonies of prairie dogs spend more time alert than rural colonies (). While scientists agree that urbanization fragments environments and directly affects animal behavior, studies of urban arthropod communities have been surprisingly rare and often limited in scope (Magle et al. 2015; Menke et al. 2011; Youngsteadt et al. 2014). Arthropods exist in multiple trophic levels in varying habitats (Crossley at al. 1973). They can also play key roles in pollination, soil aeration, and water infiltration (Barnes 1980). Ants, for example, are used as habitat indicator species for their great diversity, range of habitats, and overall ecological importance (Coovert 2005; Menke et al. 2015).

Ants are one of the most abundant insects encountered by people and inhabit a wide array of environments (Hölldobler and Wilson 1990). Many species are successful colonizers of disturbed habitats and they respond to environmental change quickly, often resulting in considerable economic impact as pests in urban and agricultural areas (Menke et al. 2015). Ants are important for seed dispersal, as they aid in creating favorable conditions for germination (Beattie & Culver 1981). Ants can even act as pollinators for plants that are considered to be self-compatible and would therefore be less affected by geitonogamy than obligate outcrossing species (Rostás et al. 2018). Ants also affect other arthropods and insects through multiple trophic levels, consuming other predators, herbivores, and plants (Rodríguez-Castañeda et al. 2016). Ants play multiple key roles in a variety of grassland and forest biomes (Menke et al. 2015; Crossley 1973); therefore, ants may contribute some of these vital roles in urban landscapes as well.

Ants in urban areas experience a different environment than ants in surrounding suburbs or rural areas. Urban ants display evidence of increased consumption of human derived foods (Penick et al. 2015). Ants living in environments dominated by pavement and concrete have greater access to human foods compared with those living in more park like environments (Penick et al. 2015). Menke et al. (2010) suggested that urban environments favor ant species that can tolerate more arid and warm environments. Prior studies in urban environments have shown areas with higher percentage of canopy (forests, greenways, and parks) tended to have a larger number of forest specialist species, in contrast to areas with less canopy cover (agricultural, business, industrial, and residential) tended to have more open habitat specialists (Menke et al. 2010; Penick et al. 2015). Penick et al. (2015) has observed greater occurrence with *Tetramorium*, typically found in urban sites compared with parks. Aside from urban areas, scientists have also been investigating ants in rural areas as well.

NaCl is limiting for ants throughout inland environments (Kaspari et al. 2008). Ant recruitment to NaCl increases in forests as distance from highways increases, demonstrating that highways act as a large source of salt in seasonal ecosystems (Kaspari et al. 2010). A larger recruitment to salt is indicative of a need for ant colonies to gather the resource. A similar pattern of salt limitation can also occur with distance from urban environments and the abundance of human enriched salt sources.

Considering urban areas tend to place more salts into the environment than rural areas, electrical conductivity (EC) should be higher in areas with dense urbanization. Salts contribute towards electrical conductivity levels in solutions (Corwin & Yemeto 2017). H_1: I predict EC will increase with proximity to human activities. Urban areas directly place additional salt into the environment. Human activity, such as increasing salt content from human foods and salt melts from winter roads, leave varying amounts of salt in the soil. Ant recruitment to salt could vary along an urban gradient moving away from a city to rural areas. H_2: I hypothesize that salt will become more limiting moving away from urban environments. H_3: I predict that ants are more likely to recruit for salt as distance from roads increases.

Methods

Electrical Conductivity

I worked with the Chicago Botanic Garden to examine soil salinity throughout the garden and if the previous use of salt application had any effect on electrical conductivity. Electrical conductivity (EC) measures the amount of electrical current that the soil could carry. Electrical current is stronger in soil with more ions, as charged ions contain higher voltage potential. When NaCl dissolves in water, Na^+ and Cl^- break from their ionic bond, placing these ions into solution. Examining the EC in an area of soil allows for the approximation of salts in a given area.

I worked with Dr. Andrea Kramer to identify and collect multiple soil samples from areas with low guest traffic, high guest traffic, near walking paths, off guest paths, and near busy roadways (Fig. 1.). In the laboratory we dissolved the soil samples with deionized water to not affect any of the preexisting ions within the soil. An EC meter was used on the solution samples to measure the electrical conductivity within the soil.

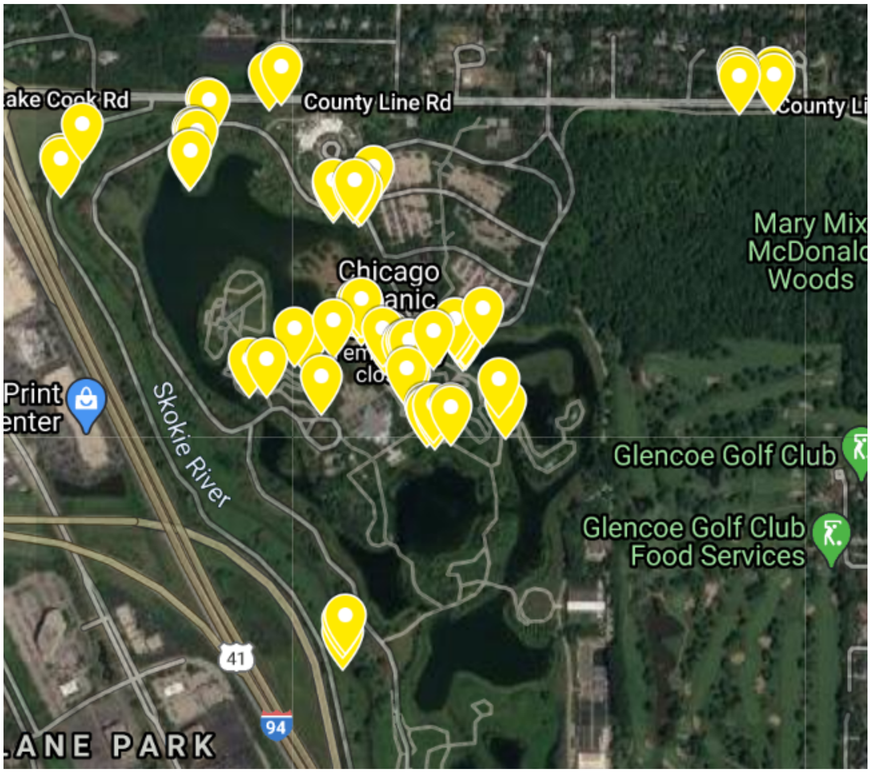

FIG. 1. Locations of soil sample sites collected throughout the Chicago Botanic Garden.

Ant community sampling

We used two transects extending 50 km from downtown Chicago to examine ant species richness and ant recruitment to nutrient baits along an urban to rural gradient (Fig. 2.). These transects were created by Lincoln Park Zoo and consisted of seventy-five sites segmented every 5 kilometers (Magle et al. 2015); the transects extend about 50 kilometers north and west from downtown Chicago. The northern transect extends from downtown Chicago and roughly follows the Des Plaines River, ending in Libertyville, Illinois. The west transect extends from downtown Chicago, following Roosevelt Road. We selected ten sites each along the north and west transects. The sites along both the north and west transects sites were separated from each other by approximately 5.5 km. We attempted to control for environment and possible confounding variables by selecting sites that contained tree cover rather than tall grasses; we focused on forested sites, rather than golf courses or city parks. The physical characteristics of each site and surrounding area changed as distance from the city increased; sites closer to the city had more buildings whereas further sites were more suburban and contained less dense concrete space. At each site we establish a sampling area of roughly 1000 square meters. Most sites were a rectangular 30m x 40m; however, due to physical limitations, some sites had varying grids such as 10m x 90m.

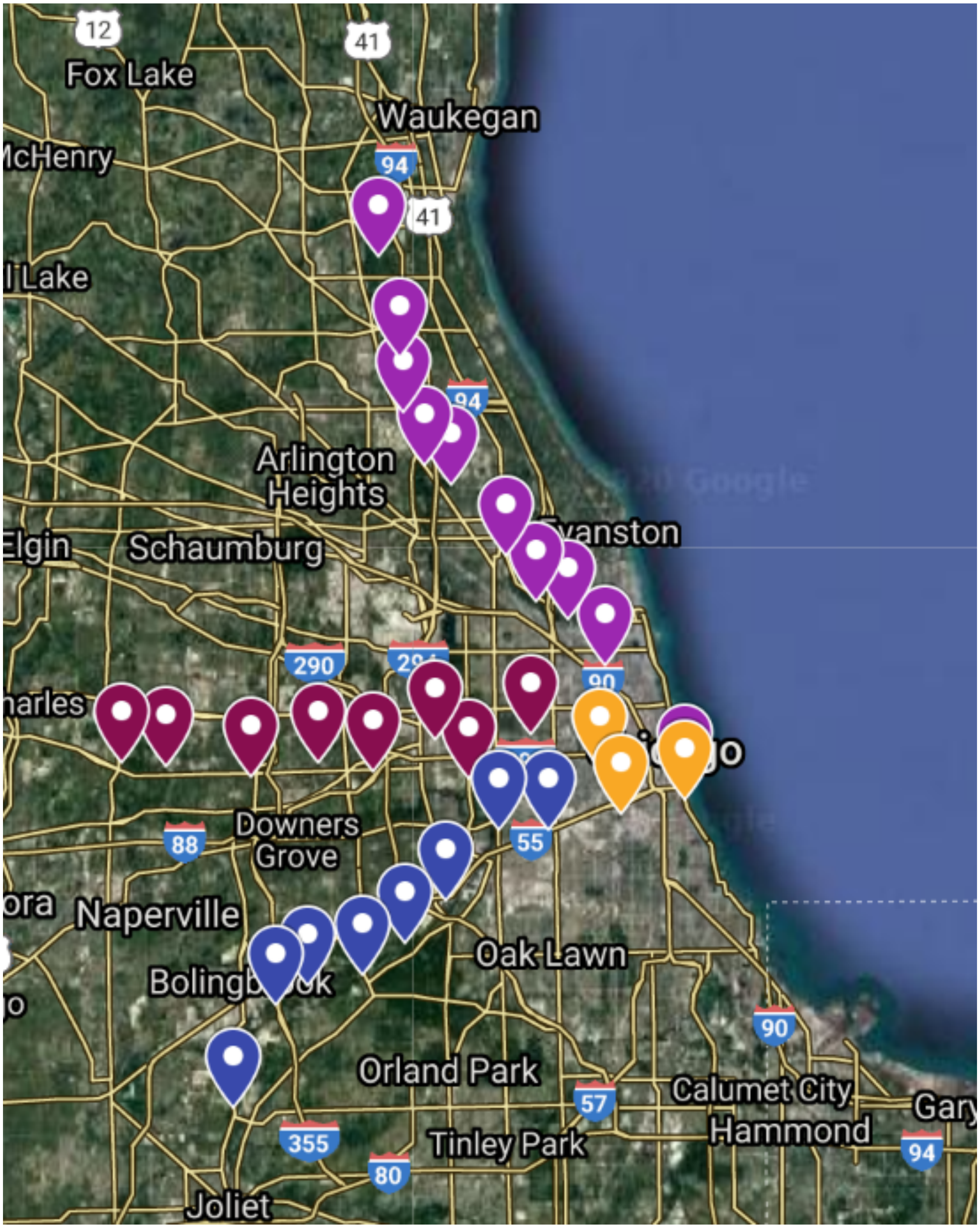

FIG. 2. Sites used along the 3 transects established by the Lincoln Park Zoo. Purple indicating the north transect, maroon indicating the west, and blue indicating the southwest (not used).

Baiting Methods

Each site was baited once between 11:00 and 17:00 hours between July 24th to October 3rd, 2019**.** Ant recruitment to baits was evaluated at each site using 3 types of baits: sugar (20%), H_2O, and NaCl (5%). Concentrations were based on previous work by Kaspari et al. (2008. While we were testing ant recruitment to salt, the use of sucrose and H_2O provided us with useful information. Sucrose recruitment in an area indicated whether ants were active and could potentially recruit to NaCl. H_2O recruitment would indicate a need for moisture; however, this was not expected as Chicago summers are relatively wet, and many sites were near water sources. We deployed ninety-nine total baits per site in an alternating order of treatments. Each site containing equal amounts (33) of the three bait types. The baits were filled with 5 milliliters of their respective solution and plugged with half a cotton ball, allowing ants a place to collect the nutrients.

While sampling a site, one vial of each type was selected in an alternating rotation, opened, and placed on the ground every 1 m. After one hour, I collected the vials, capped them to capture the ants, and returned to the laboratory for identification.

Pitfall Trapping

From June 13th to June 28th, 2019, ant incidence was evaluated by placing a series of pitfall traps at each site. At each site, the pitfall traps were dug into the ground ten meters apart from one another. This was to ensure we covered a proper area of observation, relative to the scale of ant size and their foraging distance. We used twenty 50 ml tubes per site, filled about halfway with food grade propylene glycol as a preservative and one drop of soap to break surface tension of the solution. Breaking the surface tension allows us to trap ants that could potentially stay afloat due to surface tension and walk out. Once at each site, we placed the pitfall traps into the ground with their caps on and left them for twenty-four hours. We would return to examine if any disturbances occurred to the pitfall traps, usually by people or curious animals such as coyotes and racoons. Often, we were able to place the pitfall trap back into the ground, any area that observed frequent disturbance was moved no more than 50 meters, usually away from possible pathways. Once a site was clear of disturbances, the caps were removed. The pitfall traps remained in the ground for four days, then we recapped and collected the traps.

Analyses

Soil EC from the Chicago Botanic Garden, recorded in deciSiemens per meter (dS/m), was measured for each sample and compared to the sample's $\log_{10}$ distance to the nearest confirmed salted road using a linear regression test. For baiting data, we used the percent of baits with ant recruitment. Taking the percent recruitment of a treatment per site, gives an estimate of ant activity for the particular nutrient. For pitfall trap data we used the incidence of species per site, meaning we took the number of traps that captured a particular species, divided by the total recovered pitfall traps for that site to calculate number of unique captures per trap. Simply counting the number of ants would not provide valuable information. For example, a pitfall trap could have been placed next to an existing ant colony, producing a large number of ants of a particular species in a pitfall trap. Bait recruitment and incidence were compared to increasing distance from the city and from distance to roads, using linear regressions.

Results

Hypothesis 1: electrical conductivity & road distance

The electrical conductivity (EC) measured from the Chicago Botanical Garden soil samples were examined by their distance from the nearest confirmed salted road. The EC varied throughout the sites. Sites closest to roads had the highest levels of EC, sites furthest from the road had lower EC levels. While the relationship of EC to the $\log_{10}$ distance to nearest salted road was not significant ($F_{1,62} = 3.643$, $P = 0.06204$), an overall trend suggests EC decreases with distance.

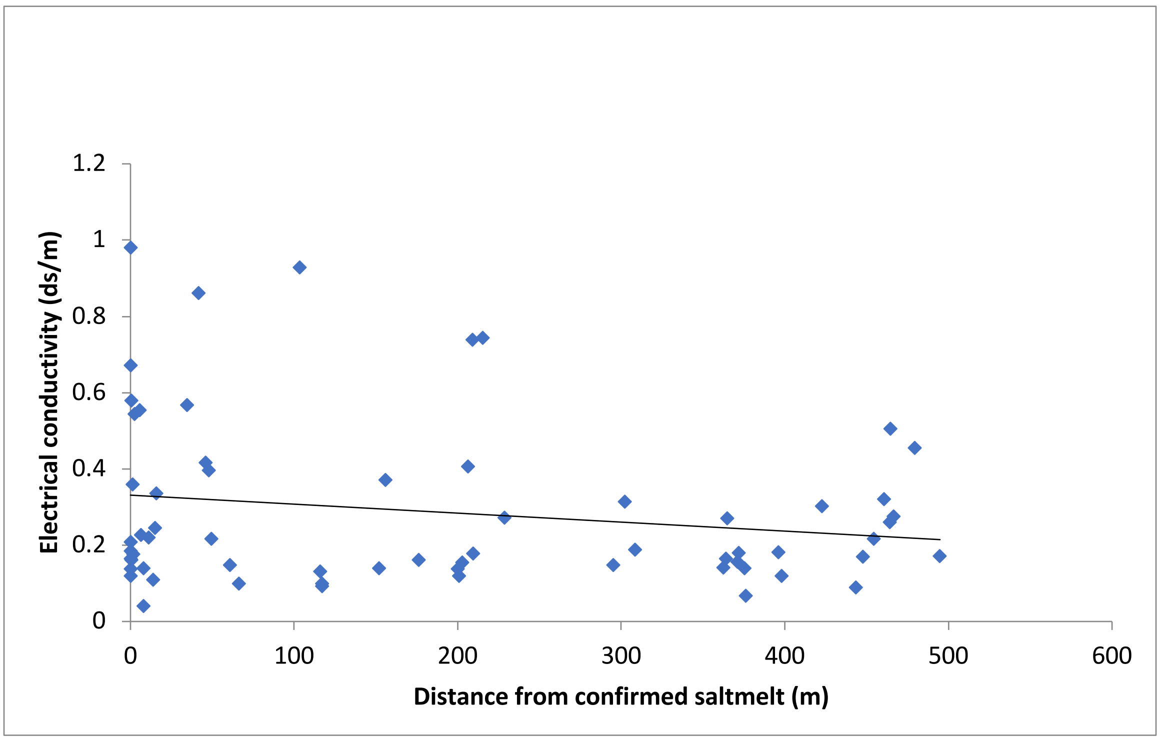

Fig. 3. The relationship between measured electrical conductivity (ds/m) and increasing distance from the closest confirmed saltmelt in meters.

Baiting and trapping

A total of 1,980 baits were set along 20 sites, across two transects. 147 of the 1,980 total baits (7.42%) showed ant recruitment. Twenty-six baits were recruited to along the north transect, while 121 were recruited to along the west transect. A total of 8 genera and 13 species were collected through baiting. While most species were not frequently found, we did find a few common species (Table. 1.). *Prenolepis imparis* was the most common per bait and per site. *Prenolepis imparis* was present in 65/147 baits and xx/xx sites. Following *Prenolepis,* the second most common genus was *Myrmica,* appearing in 55/147 baits. Other genera such as *Camponotus* and *Lasius* had much lower recruitment to baits. Percent bait recruitment by ants to NaCl and H_2O was too infrequent in both the number of sites and number of baits to analyze the respective treatments; therefore, I analyzed total percent recruitment.

One hundred forty six of 355 total pitfall traps (41.13%) captured a total of 320 ants along both transects. The traps collected a total of 15 genera and 26 species. Highest incidence, or unique captures per trap, was *Formica subsericea* (13.50%). *Tetramorium immigrans* had the 2nd highest incidence per trap (8.98%) and Myrmica detritinodis had the 3rd highest incidence (7.33%). *Brachymyrmex depilis* was found to be the most widespread, being trapped across 12 of the 20 sites.

% recruitment by Genus	Genus	Species	% recruitment by Species	Baits Recruited
1.96%	*Camponotus*	*nearcticus*	33.33%	1
		pennsylvanicus	66.67%	2
0.65%	*Formica*	*subsericea*	100.00%	1
3.92%	*Lasius*	*alienus*	50.00%	3
		neoniger	50.00%	3
1.31%	*Myrmecina*	*americana*	100.00%	2
35.29%	*Myrmica*	*detritinodis*	44.44%	24
		fracticornis	9.26%	5
		punctiventris	46.30%	25
42.48%	*Prenolepis*	*imparis*	100.00%	65
2.61%	*Tapinomoa*	*sessile*	100.00%	4
11.76%	*Temnothorax*	*ambiguus*	38.89%	7
		curvispinosus	61.11%	11
100.00%				153

TABLE. 1. The percent species and genera of ants found through baiting. The genus column shows the percentage of baits recruited to by genera. The percent species gives a percentage based off the genus. Lastly, the number of baits recruited to is shown along the farthest right column.

North Transect

The north transect had ant recruitment for H_2O, Sugar, and NaCl with 2, 23, and 1 baits, respectively. Only a single NaCl bait was recruited to by *Myrmecina americana*. Overall, the genera *Myrmica* accounted for 54% of the species collected with *Myrmica detritinodis* alone making up 29% of total bait recruitment (Fig. 4.) Some species rarely recruited to baits but were more common during pitfall trapping. *Camponotus pennsylvanicus* and the *Lasius* genera made up 26% and 16%, respectively, of the total incidence in pitfall traps along the north transect (Fig. 5.). Each, however, only accounted for 1% of bait recruitment. *Tetramorium immigrans* had 8% of the total incidence during pitfall trapping, but no recruitment to baits.

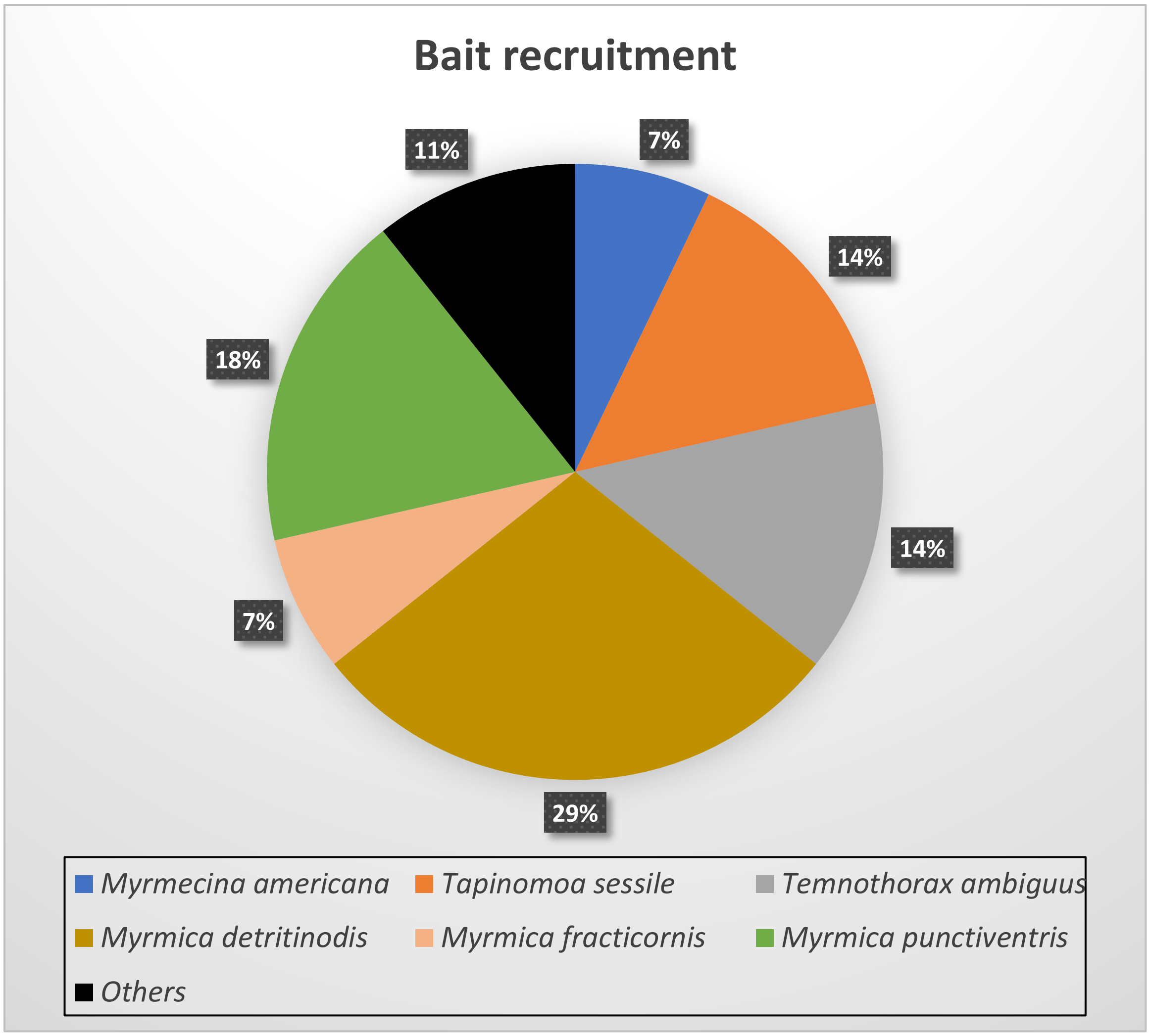

FIG. 4. The percentage of bait recruitment along the north transect. Species will low amounts of recruitment (< 7%) are in the others category.

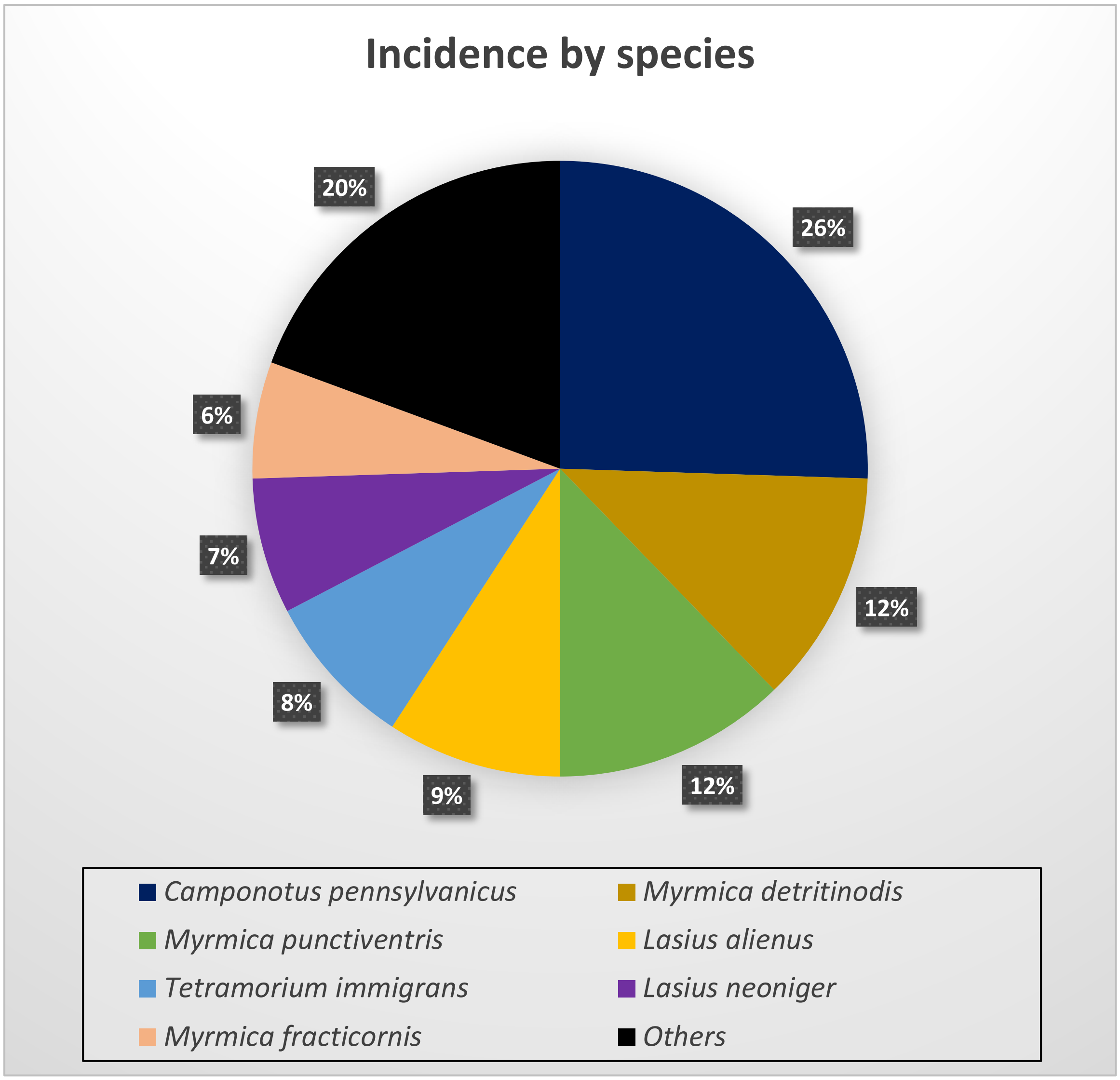

FIG. 5. The percentage of incidence through multiple species along the north transect. Species with low amounts of incidence (<6%) are in the others category.

West transect

The west transect had ant recruitment for H_2O, Sugar, and NaCl with 4, 105, and 12 baits, respectively. Of the 12 salt baits, 7 were recruited to by *Myrmica* and 5 by *Prenolepis imparis*. More than half of the total bait recruitment along the west transect was by *Prenolepis imparis* (Fig. 6.). *Myrmica* recruited to 31% of the total baits recovered. Similar to the north, the west transect trapped species that rarely, if at all, seen recruited to baits. For example, *Formica subsericea* contributed 15% of the total incidence at pitfall traps (Fig. 7), however, they only recruited to a single bait (Fig. 6.). *Tetramorium immigrans* had 10% of the total incidence (Fig. 7) during trapping, but never recruited to baits (Fig. 6).

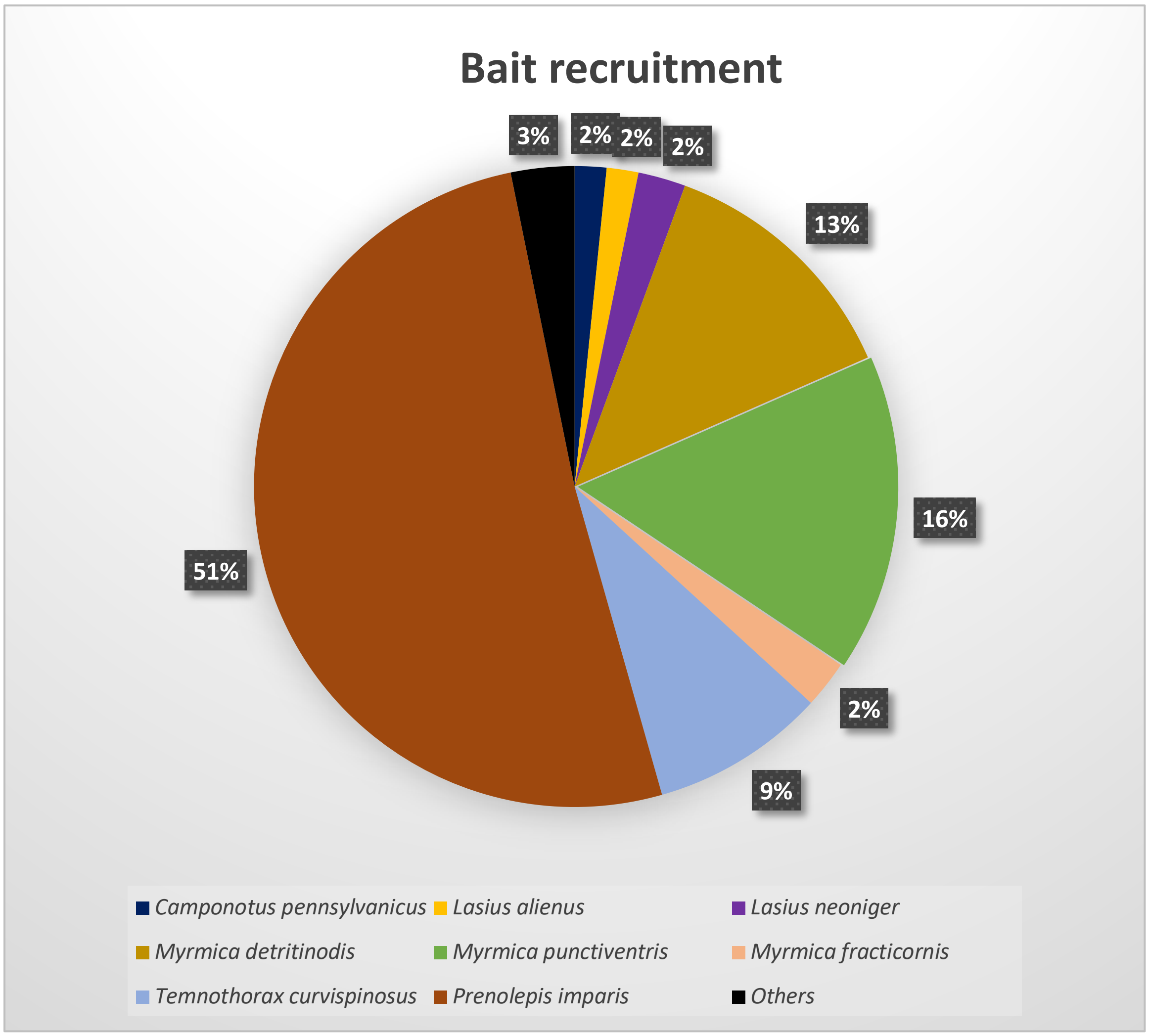

FIG. 6. The percentage of bait recruitment through multiple species along the west transect. Species will low amounts of recruitment (<2%) are in the others category.

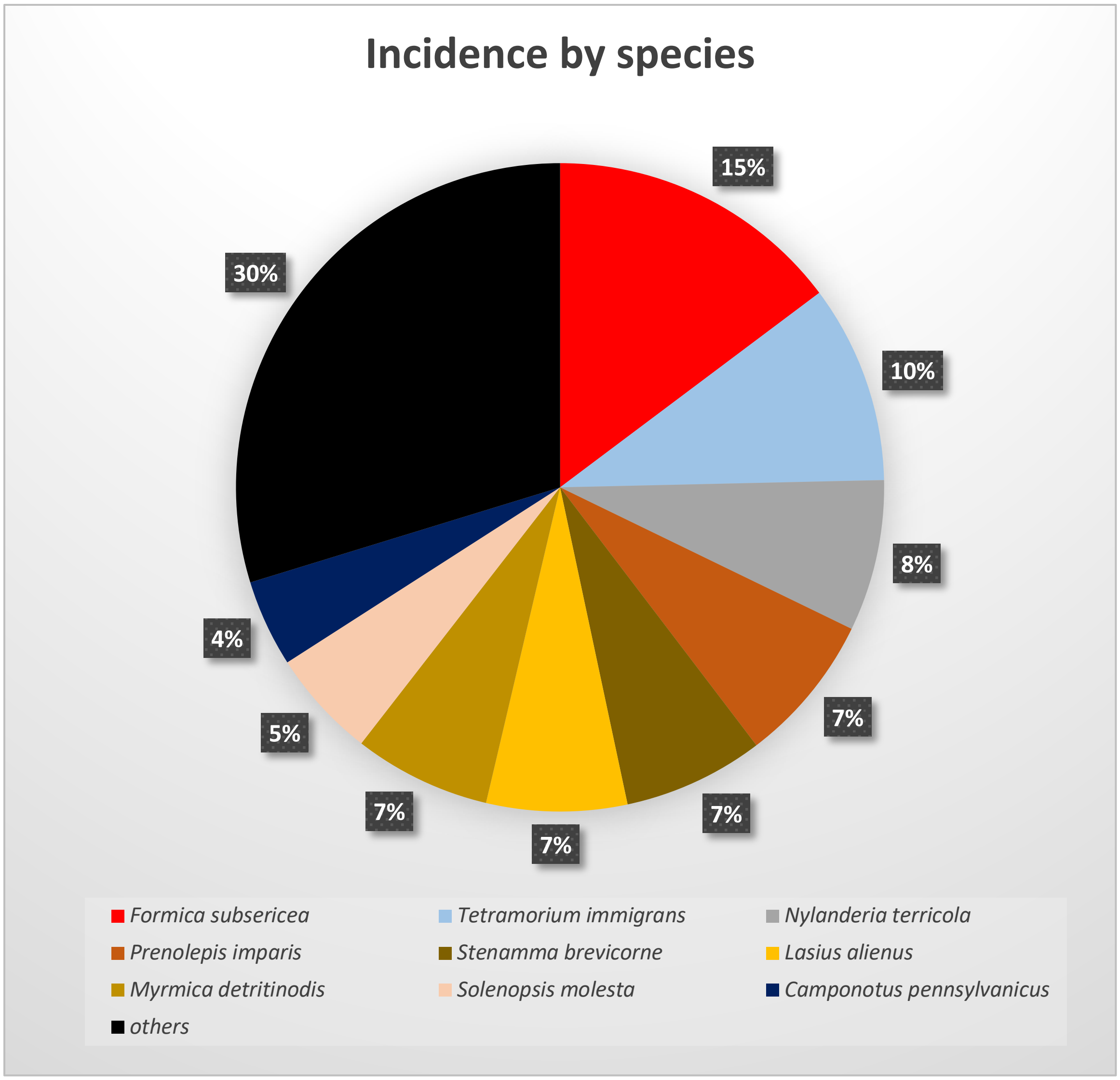

FIG. 7. The percentage of incidence through multiple species along the west transect. Species with low amounts of incidence (<5%) are in the others category.

Hypothesis 2: salts & urbanization

I first examined the relationship between percent bait recruitment and total incidence. Running a linear regression with combined data from both transects, I found there is a positive relationship between bait recruitment and incidence per pitfall trap ($F_{1,18} = 7.001$, $P = 0.0164$) (Fig. 8.). The north transect also shows a positive trend between percent bait recruitment and incidence per pitfall trap ($F_{1,8} = 5.577$, $P = 0.0459$)

(Fig. 8.). The west transect yielded no significant results ($F_{1,8}$ = 3.3005, P = 0.1068).

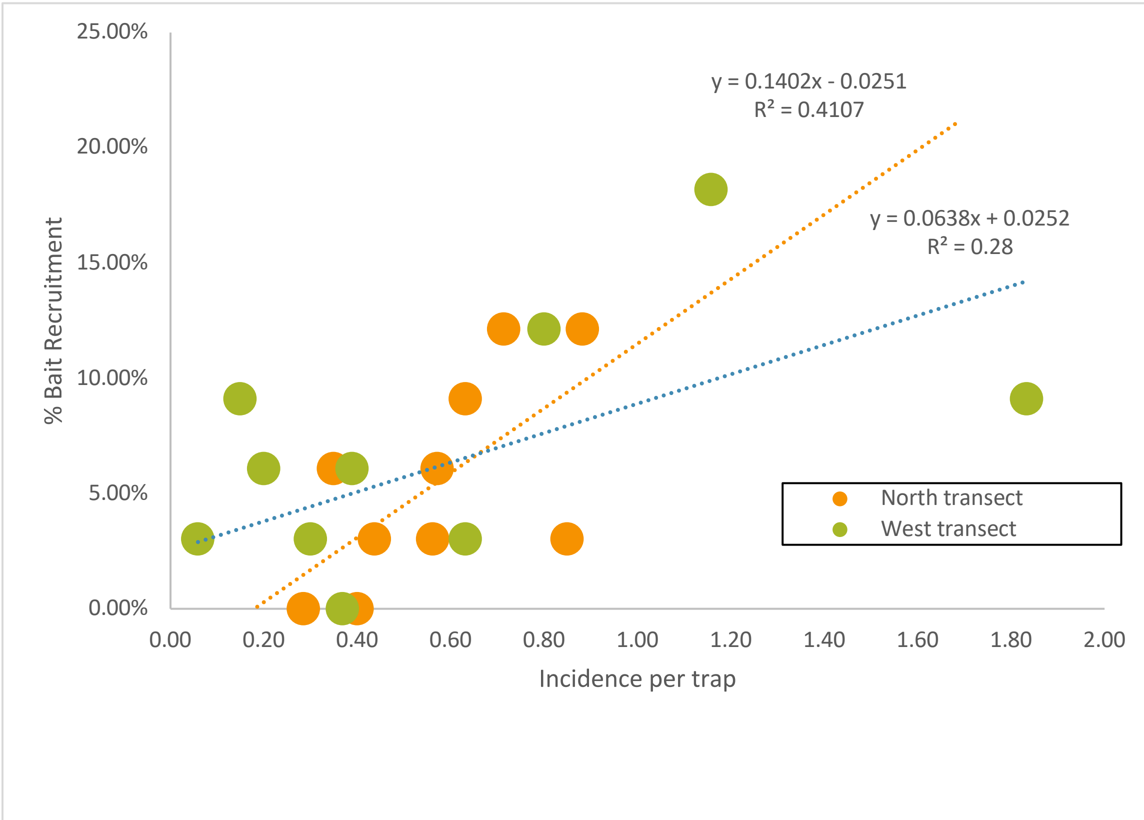

FIG. 8. The total percent bait recruitment along the y-axis, compared to the total incidence per pitfall trap on the x-axis. The blue line is the regression line between the combined data of the two transects ($F_{1,18}$ = 7.001, P = 0.0164). The orange line is the regression line for the north transect ($F_{1,8}$ = 5.577, P = 0.0459). There was no significant regression for the west transect.

After establishing a significant relationship between percent bait recruitment and total incidence per pitfall trap, I examined the relationship between percent bait recruitment and increasing distance from the city along both transects, finding no significant pattern ($F_{1,18}$ = 0.2984, P = 0.5916) (Fig. 9.). I also examined both transects separately, as both the north and west are exhibiting different patterns. The north transect begins with ant recruitment closer to the city. Low points of no recruitment are seen at 18 km and 40 km (Fig. 9.). The highest peaks of percent bait recruitment can be seen at 25 km and 52 km from the city (Fig. 9.). We see a pattern that rises and falls, however, the linear regression yielded no significant relationship between percent bait recruitment and distance from the city ($F_{1,8} = 0.064$, $P = 0.80632$). The west transect shows a single peak at 22 km, where it then decreases. This was found to be not significant ($F_{1,8} = 0.7322$, $P = 0.41705$).

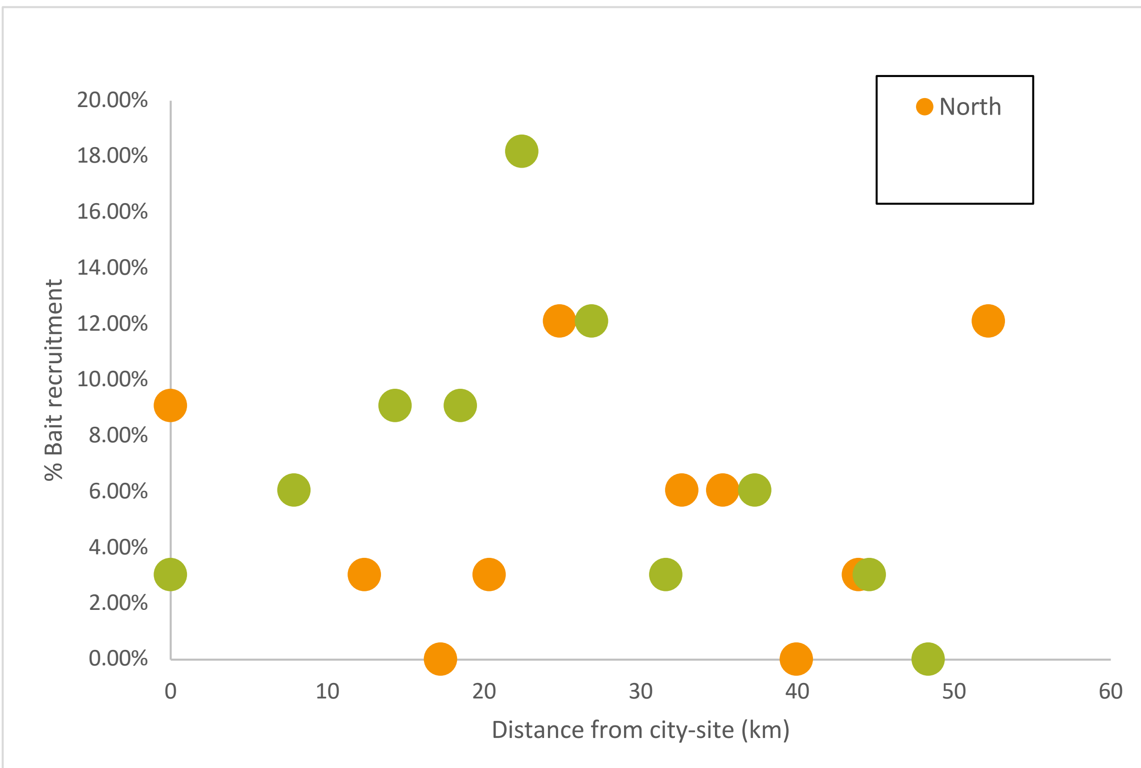

FIG. 9. Total percent bait recruitment along the y-axis, compared to the distance from each transect's city-site in kilometers on the x-axis. Combined transects ($F_{1,18} = 0.2984$, $P = 0.5916$), north transect ($F_{1,8} = 0.064$, $P = 0.80632$), west transect ($F_{1,8} = 0.7322$, $P = 0.41705$)

I also compared the total incidence per pitfall trap with increasing distance from the city. Combining the data from both transects yielded no significant results ($F_{1,18} = 1.67$, $P = 0.213$) (Fig. 10.). The north transect peaks at 25 km and 52 km. While the pattern along the north transect is not significant ($F_{1,8} = 0.3446$, $P = 0.5734$), it mimics a similar ebb and flow pattern found with the bait recruitment and increasing distance from the city (Fig. 9.). Incidence along the west transect has its highest peak at 14 km, where it then begins to decrease throughout the transect (Fig. 10.); the pattern observed along the west transect was not found to be significant ($F_{1,8}$ = 1.225, P = 0.3005).

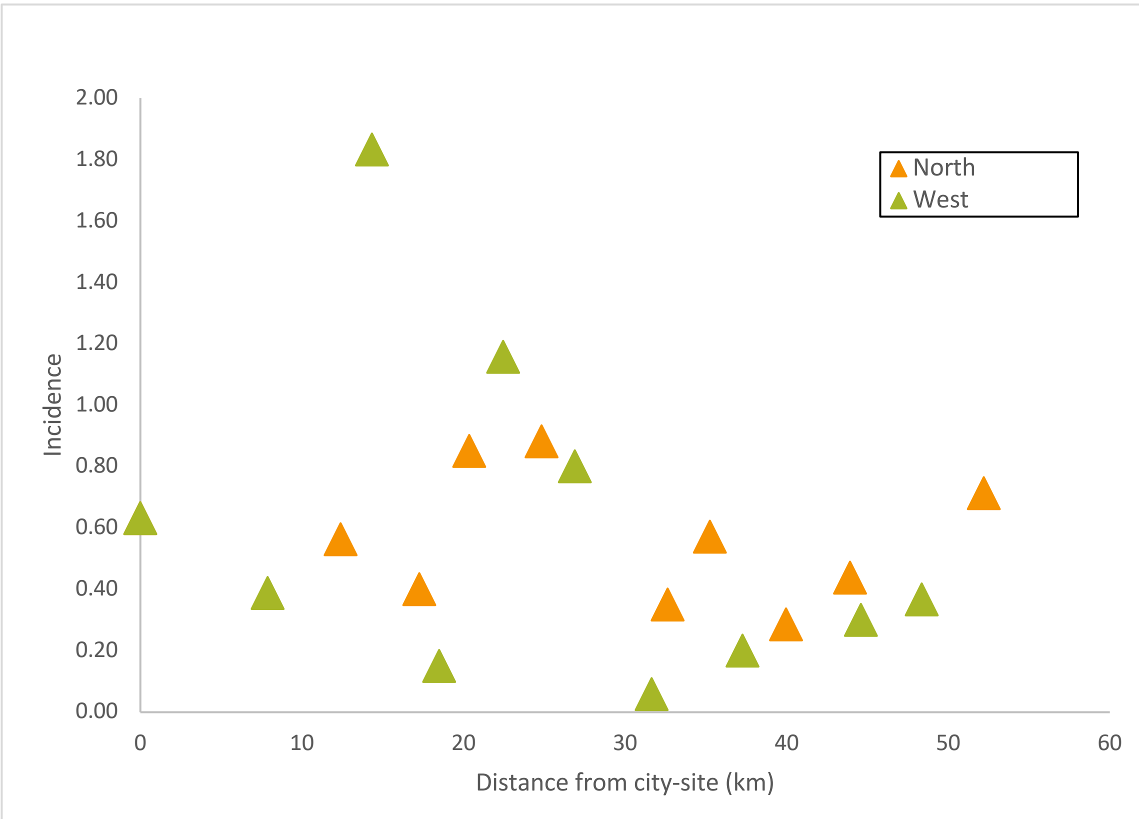

FIG. 10. The total incidence per pitfall trap along the y-axis, compared to the distance from each transect's city-site in kilometers on the x-axis. Combined transects ($F_{1,18}$ = 1.67, P = 0.213), north transect ($F_{1,8}$ = 0.3446, P = 0.5734), west transect ($F_{1,8}$ = 1.225, P = 0.3005).

Hypothesis 3: salts & road distance

There was no significant relationship between recruitment to baits and the nearest road on the combined transect data ($F_{1,18}$ = 0.275, P = 0.6064) (Fig. 11.). There was a significant positive pattern along the north transect ($F_{1,8}$ = 7.248, P = 0.0274). Along the west transect, high and low bait recruitment is seen both close to the road and further away. This was not significant ($F_{1,8} = 0.427$, $P = 0.5320$).

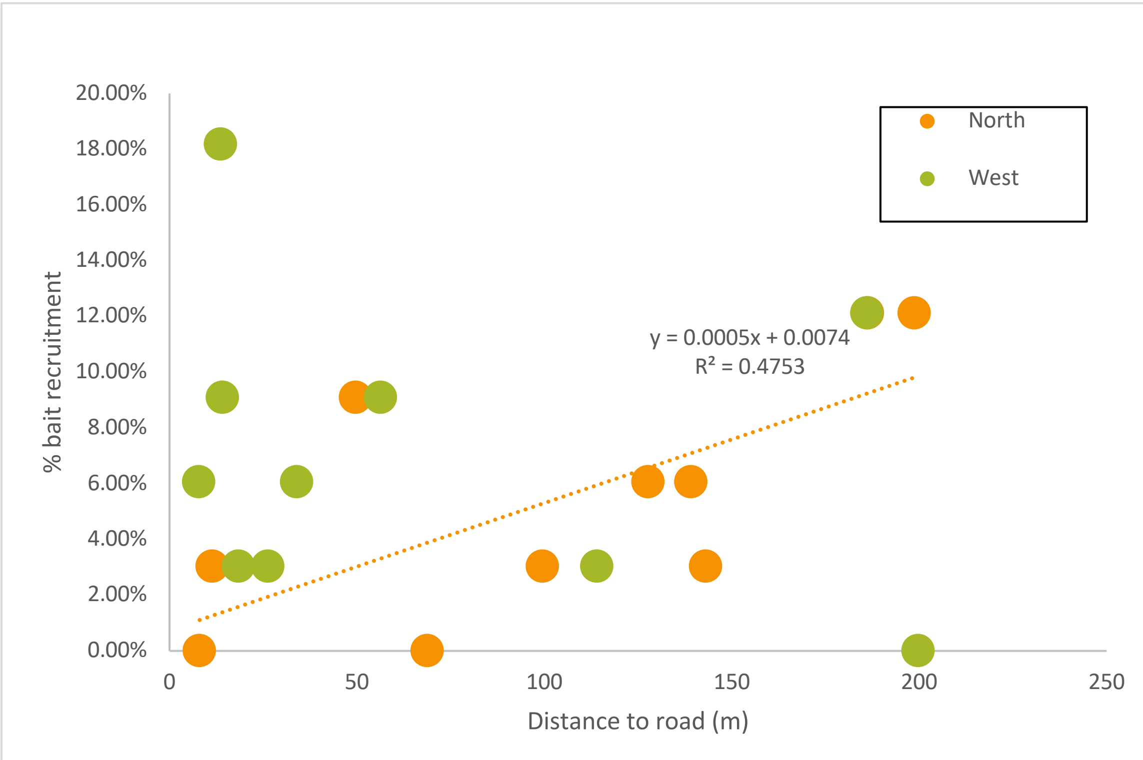

FIG. 11. The total percent bait recruitment along the y-axis, compared to the distance from a road in meters on the x-axis. Combined transects ($F_{1,18} = 0.275$, $P = 0.6064$), north transect ($F_{1,8} = 7.248$, $P = 0.0274$), west transect ($F_{1,8} = 0.427$, $P = 0.5320$).

The incidence per pitfall trap and distance from road was also analyzed with a linear regression. Examining the incidence as distance from the road increases with the combined data from both transects yields no significant trend ($F_{1,18} = 0.102$, $P = 0.7532$) (Fig. 12.). Similar to bait recruitment, the north transect appears to have an ebb and flow pattern. However, unlike bait recruitment, this apparent pattern of incidence along the north transect is not significant $F_{1,8} = 1.465$, $P = 0.2607$). Along the west transect, many of the sites are close to the road with high and low incidence. A large gap of incidence by distance is seen, except for at 100 meters. The west transect sites are seemingly not too far from roads. This was not significant ($F_{1,8} = 0.479$, $P = 0.5085$).

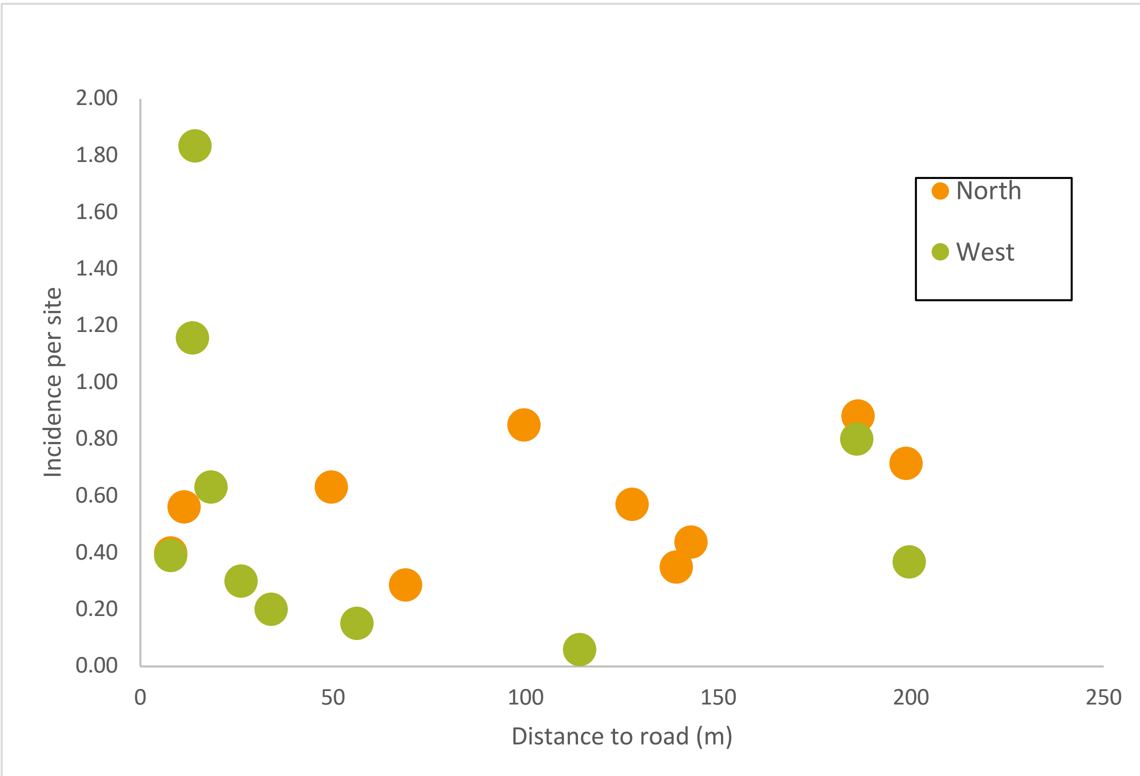

FIG. 12. Total incidence per pitfall trap along the y-axis, compared to the distance from a road in meters on the x-axis. Combined transects ($F_{1,18} = 0.102$, $P = 0.7532$), north transect ($F_{1,8} = 1.465$, $P = 0.2607$), west transect ($F_{1,8} = 0.479$, $P = 0.5085$).

Discussion

Hypothesis 1: electrical conductivity & road distance

The electrical conductivity from collected soil samples around the Chicago Botanic Garden varied greatly with distance to confirmed salted road. While we did not find a significant pattern (Fig. 3.), it is interesting that the highest EC from sampled sites occurs closer to the road than further. The farthest sites had no EC levels above 0.5 dS/ml, compared to the closer sampled areas (Fig. 3.). The closer sites with low EC could be attributed to a large brick wall that was built between Skokie highway and the western border of the Chicago Botanic Garden; this was could be effective in keeping saltmelts from easily entering the garden (Fig. 1.). The Chicago Botanic Garden is surrounded by water, an off shoot of the Skokie River corridor that runs along Chicago's northwest suburbs (Fig. 1.). After saltmelts are applied along roads, the salts quickly dissolve and are transported into rivers and lakes through leaching and runoff (Dugan et al. 2017). Saltmelt application for the de-icing of roadways has been recognized as a major source of chloride ions in groundwater, streams, rivers, and lakes throughout temperate climates in North America (Dugan et al. 2017). High levels of preexisting chloride ions and cations could already be in the water surrounding the Chicago Botanic Garden. As stated before, soil samples were collected in varying areas around the garden. While I examined the potential pattern of EC and distance from salted roads, sites nowhere near roads were close to the river system. Given that ions increase the electrical conductivity of a solution, it is highly probable that many of the soil samples collected were affected by preexisting levels of salt ions. The electrical conductivity is not exclusive to NaCl; it solely measures the voltage potential between ions, thus other salts and ionic compounds can be affecting EC.

Hypothesis 2: salts & urbanization

Our data collected on salt recruitment and distance from urbanization failed to support our hypothesis, that ant recruitment for salt would increase moving away from the city. Based on data from the bait recruitment, salt does not seem limiting along either transect. If salt were limiting, recruitment for salt baits would be higher relative to the sucrose baits. But, salt was rarely recruited to along the transects, with less than 9% of our salt baits having ant recruitment. In fact, due to salt and H_2O recruitment to baits being so low, we had to study the total bait recruitment of the three treatments combined. Additionally, there was not support for increased ant recruitment to combined baits with distance from the city either (Fig. 9.). This could suggest neither salt nor sugar is not limiting in these areas. Given that the Chicagoland area and its suburbs extends more than 100 kilometers, and that I only sampled 55 kilometers out of the city, perhaps I would not see the full effects along the urban gradient. I may need to extend further to properly observe urban and rural environments.

Examining the relationship between total bait recruitment and incidence per pitfall trap show a positive trend overall and along the north transect, however, this was not significant along the west transect. The north transect follows the Des Plaines and Skokie River moving away from the city (Fig. 2.). As observed in figures 7 & 8, as distance from the city-sites increase, the bait recruitment and incidence ebb and flow, with some areas with low incidence and no recruitment. With the north transect following water ways, these low activity sites specifically are within twenty meters of a water way. Large amounts of rain fall and flooding from these water ways could directly affect ant foraging activity (Wirth & Leal 2001). The west transect had nearly six times the amount of baits being recruited to than the north transect. The west transect does not follow a river system but follows Interstate-290 leading out of the city, converging into Roosevelt road in the west suburbs. Ants activity along the west transect would not be affected by flooding of waterways.

Hypothesis 3: salts & road distance

While a positive trend between total percent bait recruitment and distance from road was seen along the north transect, this was not the case along the west or for the combined data (Fig. 11.). Perhaps ants live on too small of a spatial scale to be impacted by examining large distances moving across the transects, being instead affected by the more proximate conditions. Ant foraging range extends between thirty to one-hundred meters away from their nest, depending in the species (Sommer & Wehner 2004). As our sites were located throughout the Chicago land area, almost none of our sites occurred over 100 meters from a road. No distance further than 200 meters was sampled in my study. While we had a few sites greater than 100 meters from the road, more data with distance would aid in examining potential patterns. Kaspari et al. (2010) found increasing distance from roads yielded higher ant recruitment for salt. The sites in more urban areas could potentially have multiple sources of anthropogenic salt rather than just the road. Salts in human foods have increased drastically, with improper waste management in urban areas, thus organisms may be consuming high salt content from human foods within their environment (National Academies of Sciences 2019; Youngsteadt 2014).

Ant Communities

The species of ants encountered in baiting and pitfall traps varied along both the north and west transect. The genus *Myrmica* was abundant at baits and traps along both transects. *Myrmica* has been observed to be an aggressive forager, quickly allowing them to recruit many workers to a food source; this allows them to monopolize sources of food quickly (Biseau et al. 1991). The genus *Camponotus* made up 28% of the incidence along the north transect but was rarely seen at baits. *Camponotus* tends to live in trees and logs. They often spend their time eating protein rich prey, mostly other insects (Rice et al. 2017). This could explain their inactivity at baits. The west transect had more differences between the species found during baiting and trapping, than the north. The west transect trapping occurred during July of 2019, with the baiting occurring during September 2019. This difference in time could account for the change in species found. For example, *Prenolepis imparis* was found at over 50% of the baits recovered, but only made up 7% of the incidence at pitfall traps along the west transect (Fig. 6). *Prenolepis imparis* is nicknamed the winter ant because they are most active in the Fall and Winter seasons (Rice et al. 2017). This could explain why *Prenolepis imparis* made up such a large amount of the baiting along the west transect. The baiting was done along the west transect during September, while the pitfall trapping was done during June 2019. This timing difference can also explain the differences of species found between baiting and pitfall trapping along the west transect (Fig. 6. & Fig. 7.). Lastly, the traps were left for several days to collect species, whereas the baits were only left for an hour. The pitfall traps have more time to collect ants that live in different areas, compared to the baits that may not observe recruitment from ants that live in trees or the ground

Environmental conditions may have influenced patterns in species sampling. Chicago's precipitation during 2019 was the 3rd highest ever recorded since 1871, with 49.54 inches of rain (N.O.A.A. 2020). The average rainfall of July in Chicago is 3.70 inches of rain. The rainfall during 2019 was 3.94 inches (N.O.A.A. 2020). This higher amount of rainfall could have inhibited ant activity along the north transect, in which the sites run along water systems (Wirth & Leal 2001). The west transect received 7.61 inches of rainfall in September when baiting occurred, with the average precipitation in September being 3.21 inches. While the west transect does not lay along water systems, the environment was likely affected from the large amount of precipitation.

Further research

Data on electrical conductivity was measured at the Chicago Botanic Garden. Future research along the transects can collect soil samples from the sites to measure EC. This would allow comparison of incidence and bait recruitment to the EC of the soil for each site. The electric conductivity does not directly measure NaCl; isolating ions in biomass may be a better measurement of salt ions (Penick et al. 2015). The EC can also be measured with increasing distance to the river systems. The data collected could be used to assess the soil salinity along the different transects.

To investigate ant community composition, baits and traps can be set along northern areas that are further from the water systems. This could attempt to mitigate confounding variables from the waterways, such as additional ions from saturated sources (Dugan et al. 2017). Sites further than the 55 km length of each transect can be explored as well to perhaps see more of a difference along the urban gradient. Additionally, sites further than 100 m from a road can be investigated. This may require data to be collected from areas extending further than the previously established Lincoln Park Zoo transects. Most interestingly, however, my study examined the north and west transect. Examining Lincoln Park Zoo's southwest transect could add insight to the data already found by comparing baiting and incidence along anther waterway. This would allow a comparison of ant communities and how they may vary to north and west transect. Both the southwest and north transect follow waterways.

Conclusion

Expanding urbanization can lead to changes in salinity levels throughout the ecosystem. Here, I attempted to determine how ants responded to salt availability along an urban to rural gradient. While my study was unable to directly examine salinity and its effect on ant recruitment, I was able to find data suggesting recruitment and ant communities may be influenced by waterways and surrounding conditions. Future empirical studies should be conducted, exploring input of salt into the environment and its effect on organismal behavior. I studied ants because of the multiple roles they play in ecosystems. Examining the effects of anthropogenic salts on arthropods can show patterns on a small spatial scale and could then affect other trophic levels. NaCl in ecosystems can play the role of a toxin and/or a limiting nutrient (Kronzucker 2013). Exploring the potential patterns and consequences on organisms is crucial for maintaining the urban environment that we share.

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